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Optimization of non-invasive optical methods for quality control of wine grapes

Giovanni Agati¹, Elisa Fierini, Patrizia Pinelli, Annalisa Romani

Department of Pharmaceutical Sciences, University of Florence, Via Ugo Schiff 6, 50019 Sesto F.no (Florence - Italy)

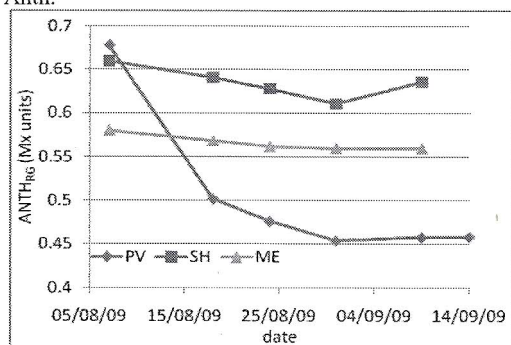
¹IFAC CNR, Via Madonna del Piano 10, 50019 Sesto F.no (Florence -Italy)

Abstract. Accumulation of anthocyanins (Anth) on whole winegrape (*Vitis vinifera* L.) bunches attached to the vine was followed non-destructively using a fluorescence-based sensor during the 2009 season in the Central Italy (Latina, Lazio). Measurements were performed weekly from véraison to harvest on Merlot, Shiraz and Petit Verdot cultivars. The ANTH_{RG} index showed a different time evolution and Anth accumulation for the 3 cultivars. The data obtained from the sensor have been compared with the spectrophotometric analyses in order to assess the anthocyanoside content. The fluorescence-based sensor used in the present study represents a rapid and non-invasive tool for the assessment of grape phenolic maturity and quality in vineyards.

Introduction. The wine industry needs a more in-depth knowledge of product for the characterization and the definition of organoleptic parameters, in particular, winegrape phenolic maturity is a fundamental parameter to be controlled for the production of high-quality wine. Grape phenolic maturity is usually determined by destructive laboratory analysis [1], which are time-consuming and require an accurate sampling approach to be representative of the vineyard. The object of the present study has been the assessment of the anthocyanin (Anth) content of three grape cultivars sampled from "Casale del Giglio" (Latina, Lazio) winery, employing an innovative and non invasive optical method using a portable LED-based sensor recently developed and tested on grape bunches in the laboratory [2]. The method is based on the screening of fruit chlorophyll fluorescence and allows the Anth contents of intact berry skin to be measured [3]. With increasing Anth content, less excitation light was transmitted to the deeper chlorophyll layers, and thus the chlorophyll fluorescence signal decreased proportionally.

Materials and Methods. The Multiplex 3 (Mx) (FORCE-A, Orsay, France) was an hand-held battery-operated optical sensor consisting of 4 excitation LED sources in the UV-A (375 nm), blue (470 nm), green (516 nm) and red (635 nm) and 3 detection channels in the blue-green, red and far-red spectral regions [2]. The ANTH_{RG} index was calculated as $\log(F_R/F_G)$, where F_R and F_G are the chlorophyll fluorescence excited with red and green light, respectively. The colorimeter Maselli MT01 (Maselli Misure S.p.A., Parma, Italy) and the spectrophotometer DAD/UV-Vis (Cary 50, Varian) have been used to assess the content of anthocyanosides by destructive analyses using a calibration curve of malvidin 3-O-glucoside.

Results and Discussion. The fluorescence-based sensor has been applied directly in the field to follow the time evolution of Anth accumulation in the same plots of the vineyard. Grape bunches attached to the vine from Merlot (ME), Petit Verdot (PV) and Shiraz (SH) cultivars were monitored from complete véraison to harvest (Aug 7th – Sep 14th, 2009). For each measuring date, samples of grape berries from the 3 cultivars were collected and used for the destructive extraction and spectrophotometric analysis of Anth.



The time course of the ANTH_{RG} index derived by Mx measurements *in situ* is reported in Figure 1 for the investigated cultivars. The index decreases exponentially starting from complete véraison, with different time evolution and Anth accumulation for the 3 cultivars. PV maturation curve was delayed with respect to ME and SH, but reached the highest concentration of Anth (lowest ANTH_{RG} values).

Figure 1. Time course of the anthocyanin index ($ANTH_{RG}$) measured in situ by the multiplex sensor. In order to calibrate the Mx sensor for the Anth content, for each date 3-6 different bunches per cultivar were randomly collected. Single bunches were measured *in situ* by the Mx, then, samples of 150 berries were randomly collected and processed for Anth extraction and quantification by spectrophotometric and colorimetric methods. Weighed berries were crushed inside the Maselli device and the slurry was measured within fifteen minutes. Five grams slurry aliquots were then extracted (overnight) in 45 ml EtOH:H₂O:HCl (73:27:1) pH 1.8 and measured by the spectrophotometer after filtration. Total anthocyanins were expressed as malvidin-3-O-glucoside. The data obtained from the sensor have been compared with the results carried out by the destructive analyses. Figure 2 reports the correlation between Mx results and the quantitative data obtained using the colorimeter technique for all the cultivars.

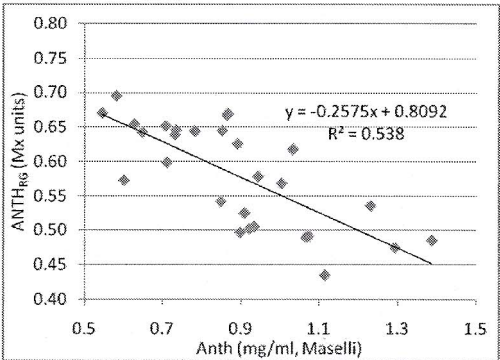


Figure 2. Comparison between the mean $ANTH_{RG}$ index from the sampled bunches and the quantitative results obtained by colorimeter method. The calibration curve for the assessment of Anth content by the Mx sensor in the investigated cultivars using the spectrophotometric destructive analysis is presented in Figure 3. The best fitting ($r^2 = 0.82$) has been obtained with an exponential curve [$ANTH_{RG} = 0.38 + 13.22 \cdot \text{EXP}(-2.41 \cdot \text{Anth})$]. By inverting this function, and using the Mx index values measured in the vineyard, it is possible to obtain the predicted Anth concentration per cv per plot. At harvest, Anth was 2.1, 1.8 and 1.6 g/kg for PV, ME and SH, respectively.

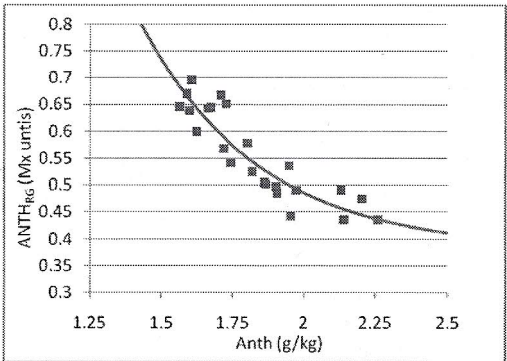


Figure 3. Calibration curve for the $ANTH_{RG}$ index measured on red berries for the investigated cultivars. According to our results, the proposed Mx screening method can represent a new rapid and non-invasive tool for the assessment of phenolic maturity in red grape vineyards.

References

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